mycins under similar conditions. This is not surprising if it is assumed that an essentially free carbonyl group is necessary for the rearrangement. The aldehyde group in the streptomycin molecule at a *p*H of 12.5 or less is apparently partially bound. Thus, in solutions of these *p*H's, every streptomycin molecule does not produce an essentially free aldehyde group which is capable of rearrangement. Consequently, the rate of decomposition of streptomycin should be slower than the rearrangement of α-hydroxyisobutyraldehyde where all the aldehyde groups are essentially free.

Acknowledgment.—The authors wish to thank the Heyden Chemical Corporation for supplying all of the streptomycin that was used in this investigation.

Summary

1. A study has been made of the polarographic reduction of streptomycin and mannosidostreptomycin over the *p*H range of 4.2 to 14.

2. The rate of decomposition of streptomycin has been followed polarographically in various alkaline phosphate and borate buffers and in 1 *N* sodium hydroxide.

3. In the *p*H range 9.6 to 14, the rate of decomposition of both streptomycins has been shown to be identical in phosphate buffers and to follow a first order reaction. The rate of decomposition for these compounds differs slightly in borate buffers. The energy of activation, free energy of activation and entropy of activation were calculated for this reaction.

4. The rate of rearrangement of α-hydroxyisobutyraldehyde to form acetoin was measured and compared with the rate of decomposition of streptomycin. Both of these reactions are first order, require essentially the same energy of activation, and show the same *p*H dependence. It was concluded that the rate determining step in the alkaline decomposition of streptomycin is probably the rearrangement of the streptose moiety to form a six-membered ring.

5. Because of the great similarity of the two streptomycins in their polarographic behavior and in their rates of decomposition, it is unlikely that a simple polarographic technique can be used to determine these compounds in the presence of each other.

PRINCETON, NEW JERSEY RECEIVED AUGUST 7, 1950

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF MERCK & CO., INC.]

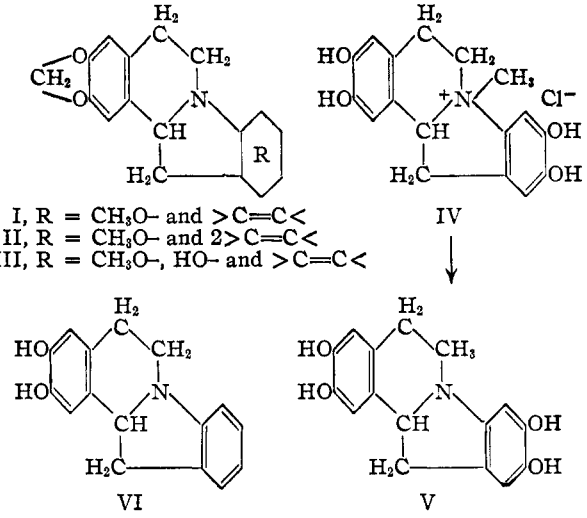
Erythrina Alkaloids. XVIII. Studies on the Structure of Erysopine, Erysodine, Erysovine and Erythraline

BY KARL FOLKERS, FRANK KONIUSZY AND JOHN SHAVEL, JR.

It was established[1] that erysopine, erysodine and erysovine have the same four-nuclei ring system and that each has three oxygen atoms at the same positions on the ring system. However, these three alkaloids differ in the number and position of O-methyl groups. These deductions are based on analytical, hydrogenation, and methylation experiments, and provided a basis for the new Hofmann degradation and other reactions which are described herein.

The structures[2] of erythramine (I), erythraline (II) and erythratine (III) also provided a basis for further work on erysopine, erysodine and erysovine since all of these alkaloids seemed to have related structures. The conversion of laudanosoline into dehydrolaudanosoline chloride (IV) and then into 2,3,11,12-tetrahydroxydibenzotetrahydropyrrocoline (V) has been discovered by both Robinson[3] and Schöpf[4] and their coworkers. The properties of the pyrrocoline derivative (V) and the Hofmann and Emde degradations of it were promising precedents for these new Erythrina alkaloid studies.

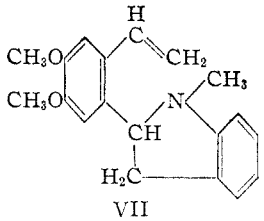
Erysodine and erysovine have the formula $C_{18}H_{21}NO_3$, and have two methoxyl groups and one hydroxyl group; erysopine has the formula $C_{17}H_{19}NO_3$, and one methoxyl group and two



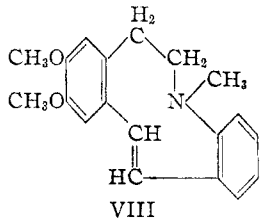
hydroxyl groups.[5] Each of these alkaloids has two double bonds in addition to the benzenoid nucleus and yields tetrahydro derivatives.[1,6] It was desirable to aromatize the ring containing the two double bonds before attempting a Hofmann degradation. This aromatization was accomplished by treatment of the alkaloid with hydrobromic acid which resulted in a decrease of CH_4O in composition. The reaction of erysopine with

(1) Koniuszy, Wiley and Folkers, THIS JOURNAL, **71**, 875 (1949).
(2) Folkers, Koniuszy and Shavel, *ibid.*, **64**, 2146 (1942).
(3) Robinson and Sugasawa, *J. Chem. Soc.*, 789 (1932).
(4) Schöpf and Thierfelder, *Ann.*, **497**, 22 (1932).
(5) Folkers and Koniuszy, THIS JOURNAL, **62**, 1677 (1940).
(6) Folkers and Koniuszy, *ibid.*, **72**, 1832 (1950).

47% hydrobromic acid at 110–120° gave a crystalline product, $C_{16}H_{15}NO_2$, which is designated apoerysopine and is assigned structure VI. As could now be predicted, apoerysopine was also obtained from the reaction of erysovine with hydrobromic acid. Apoerysopine reacts with ferric chloride



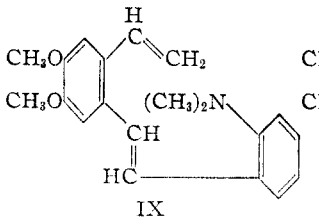
VII



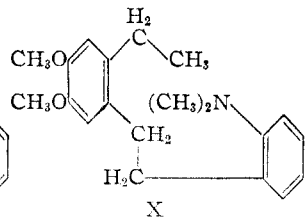
VIII

in water to give a purple color which is characteristic of the two phenolic hydroxyl groups in ortho position. It gives an initial blue-green color with Ehrlich reagent and then the color becomes deep purple on standing, due no doubt to reactions, including oxidation, of the pyrroline nucleus.

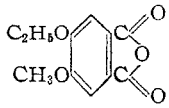
The first-stage Hofmann degradation of apoerysopine by methylation with dimethyl sulfate and ring scission with 30% potassium hydroxide solution gave a crystalline product, $C_{19}H_{21}NO_2$, which probably has structure VII or possibly VIII, and is designated *des*-methylapoerysotrine.[7] When a second-stage Hofmann degradation was applied to *des*-methylapoerysotrine, a product was obtained which melted at 97.5–98° and has the composition $C_{20}H_{23}NO_2$; it is designated *des*-dimethylapoerysotrine (IX). The mother liquor from *des*-dimethylapoerysotrine was retreated with dimethyl sulfate and potassium hydroxide. Another product, $C_{20}H_{23}NO_2$, was obtained which is possibly a geometric isomer of *des*-dimethylapoerysotrine. A third-stage degradation was attempted upon *des*-dimethylapoerysotrine (IX), but no trimethylamine was formed and the starting material was recovered. *des*-Dimethylapoerysotrine was hydrogenated in ethanol with a platinum catalyst to give



IX



X



XI

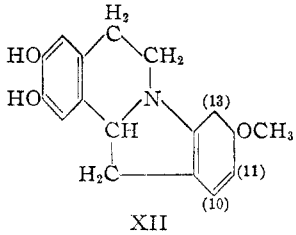
tetrahydro-*des*-dimethylapoerysotrine (X), and an Emde degradation was attempted once on this compound. However, a nitrogen-free product was not obtained. Emde degradations on similar compounds either failed or gave a low yield of nitrogen-free product.[3]

Oxidation of *des*-dimethylapoerysotrine to give N-dimethylanthranilic acid would be of interest. However, it appears that N-dimethylanthranilic acid would probably not be stable under suitable conditions for oxidation of the ethylenic groups, and it would be further oxidized.[8] The oxidation product of N-dimethylanthranilic acid is not well characterized. Hence, N-dimethylanthranilic acid was synthesized, by a new modification, for use in "model oxidation reactions." Unfortunately, there was no opportunity to complete a "model oxidation" and apply it to *des*-dimethylapoerysotrine.

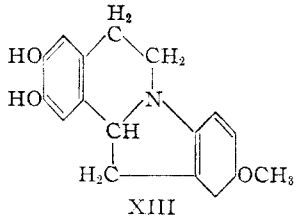
When erysodine was added to molten potassium hydroxide, the fusion reaction yielded indole in a rather good yield. Indole was also obtained[2] from erythraline by alkali fusion.

Treatment of erysodine with diethyl sulfate and oxidation of the product with potassium permanganate gave 4-ethoxy-5-methoxyphthalic anhydride (XI). Similar degradation of erysovine and "erysocine" also gave 4-ethoxy-5-methoxyphthalic anhydride. The phthalic derivative was also characterized as 4-ethoxy-N-ethyl-5-methoxyphthalimide. This phthalic anhydride derivative (XI), which was obtained from the alkaloids, and an authentic specimen obtained from the oxidation of the 7-ethoxy-6-methoxy-3,4-dihydroisoquinoline were identical.

Structures XII (somewhat preferred) and XIII may now be considered for erysopine with the thought that the methoxyl group at position 10 or 13 is not excluded. Erysodine and erysovine, on

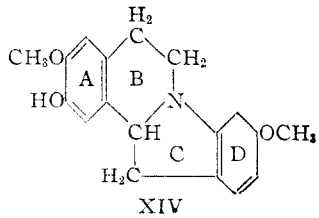


XII

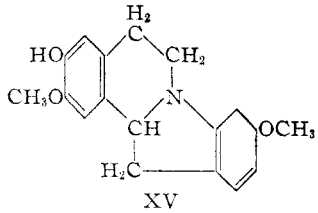


XIII

the basis of structure XII for erysopine, will have structures XIV and XV, but the relative position of the methoxyl group is unknown. Alternative structures for erysodine and erysovine, based on XIII, are obvious.



XIV

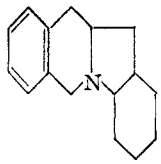


XV

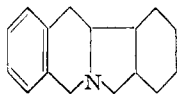
Ring system (XVI) is excluded on the basis of the composition of *des*-dimethylapoerysotrine, and the system XVII is excluded on the basis of the formation of indole.

(7) This name is based upon the erysopine–erysotrine nomenclature and the Willstätter convention for the products of exhaustive methylation.

(8) Willstätter and Kahn, *Ber.*, **37**, 409 (1904).

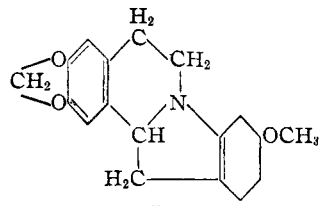


XVI

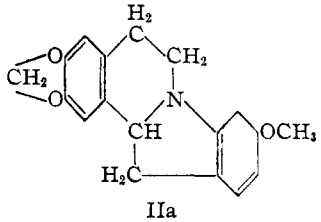


XVII

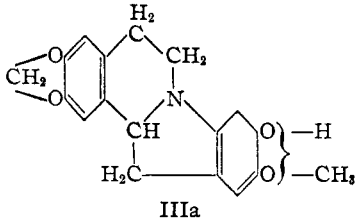
Proof that erythraline has the same four-nuclei ring system as erysopine and related eryso-alkaloids resulted from the degradation of erythraline to apoerysopine (VI) by reaction with hydrobromic acid. Consequently, structure Ia for erythramine, structure IIa for erythraline and structure IIIa for erythratine may now be considered for these alkaloids.



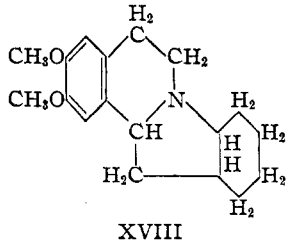
Ia



IIa



IIIa



XVIII

Prelog, Wiesner, Khorana and Kenner have recently described some reactions and degradations of *Erythrina* alkaloids. Structures Ia and IIa can account for the conversion[9] of erythraline to erythramine by hydrogenation, and structures XIV or XV can account for the formation of dihydroerysodine.[9] The differences observed between the products,[9] $C_{18}H_{25}NO_2$, which were derived from erythraline and erysodine, appear to be due to steroisomerism and cannot be due to a difference in the carbon–nitrogen skeleton of erythraline and erysodine, because of our conversion of erythraline to apoerysopine. Structure XVIII accounts for the products, $C_{18}H_{25}NO_2$.

(9) Prelog, Wiesner, Khorana and Kenner, *Helv. Chim. Acta*, **32**, 453 (1949).

Labriola, Berinzaghi and Deulofeu[10] have prepared crystalline erysotrine which was degraded to the imide of hemipinic acid by oxidation, and concluded that a part of the structure of the free alkaloids coincides with that of the liberated alkaloids.

Synthesis[11] in the series of *Erythrina* alkaloids by Wiesner, Clarke and Kairys has yielded a compound (XVIII), which on the basis of the respective degradative and synthetic studies is a racemate of "hexahydroapoerysotrine."

Experimental

Conversion of Erysopine into Apoerysopine.—Four grams of erysopine was dissolved in 36 ml. of 48% hydrobromic acid and the mixture was heated in a sealed tube at 110–120° for 2 hours. After cooling, the purple colored solution was diluted with 200 ml. of water and the solution was extracted ten times with 50-ml. portions of chloroform. Distillation of the chloroform left a meager residue which was discarded. The aqueous solution was then made alkaline by adding sodium bicarbonate and then it was extracted again with ten 50-ml. portions of chloroform. Concentration of the extract by distillation to a volume of about 50 ml. yielded 1.2 g. of gray-colored crystals. One recrystallization of this material from ethanol and two recrystallizations from chloroform yielded 624 mg. of apoerysopine, m.p. 169.5–170.5°. The addition of ferric chloride to an alcoholic solution of apoerysopine gives a deep purple color. The compound was dried at 78° *in vacuo* before the analyses.

Anal. Calcd. for $C_{16}H_{15}NO_2$: C, 75.87; H, 5.93; N, 5.53. Found: C, 75.80; H, 5.95; N, 5.65; $-OCH_3$, none.

Conversion of Erysovine into Apoerysopine.—Each of two 4-g. samples of erysovine was dissolved in 36 ml. of 48% hydrobromic acid, and the two solutions were heated in sealed tubes at 110–120° for 2 hours. When cool, the solutions were diluted with 200 ml. of water, combined, and washed with five 50-ml. portions of chloroform. The aqueous solution was treated with sodium bicarbonate to *p*H 8, and extracted with ten 50-ml. portions of chloroform; distillation of this solvent extract gave 3.4 g. of dark partially crystalline residue. A solution of this residue in 50 ml. of methanol was treated with 0.5 g. of norite, and the filtrate was diluted with petroleum ether to incipient turbidity. This solvent mixture was concentrated to a volume of 25 ml. whereupon crystallization took place. The apoerysopine weighed 3.0 g. and melted at 167–170°; after recrystallization from methanol–petroleum ether, it melted at 169–170°.

Degradation of Erythraline to Apoerysopine.—A 104-mg. sample of erythraline hydriodide was dissolved in 5 ml. of 48% hydrobromic acid and the solution was heated in a sealed tube at 110–120° for 2 hours. After cooling, the solution was diluted with 25 ml. of water, and washed with six 25-ml. portions of chloroform. The solution was then made alkaline with sodium bicarbonate and extracted with six 25-ml. portions of chloroform. Concentration of the chloroform extract from the alkaline solution yielded 41 mg. of a brownish colored gum. This residue was dissolved in 2 ml. of methanol, and, after adding 2 ml. of petroleum–ether, the solution was filtered and allowed to stand 3 days at room temperature. The yield of crystals, which melted at 168–170°, was 14 mg. This material was placed in a micro-sublimation tube and heated at 170–175° and 0.1 mm. pressure. The white crystalline sublimate melted at 170–171°, and when this material was mixed with apoerysopine, there was no depression in the melting point of the mixture.

First Stage Hofmann Degradation of Apoerysopine to *des*-Methylapoerysotrine.—Five hundred milligrams of apoerysopine was dissolved in 5 ml. of 30% potassium hydroxide solution and 3 ml. of dimethyl sulfate was added. The mixture was shaken for 15 minutes and, after adding 5 ml. of 30% potassium hydroxide solution, two 3-ml. portions of dimethyl sulfate were added at intervals of 15 minutes and with occasional shaking of the mixture. After this

(10) Labriola, Berinzaghi and Deulofeu, *Cienciae Investigation*, **5**, 349 (1949).

(11) Wiesner, Clarke and Kairys, *Can. J. Research*, **28**, 234 (1950).

methylation procedure, 30 ml. of 30% potassium hydroxide solution was added and the mixture was heated on the steam-bath for 30 minutes. A light-colored oil separated and, after cooling, was extracted with ether. Distillation of the ether yielded 293 mg. of residue; the gummy base crystallized from ethanol. After two recrystallizations, 83 mg. of *des*-methylapoerysotrine was obtained; m.p. 72–73°. The compound was dried at 25° *in vacuo* before the analyses.

Anal. Calcd. for $C_{19}H_{21}NO_2$: C, 77.26; H, 7.17; N, 4.74; $2CH_3O$—, 21.01; CH_3N=, 5.09. Found: C, 77.46; H, 7.37; N, 4.75; CH_3O—, 18.84; CH_3N=, 7.78.

Second Stage Hofmann Degradation of *des*-Methylapoerysotrine to *des*-Dimethylapoerysotrine.—Five grams of *des*-methylapoerysotrine was dissolved in 100 ml. of benzene and 25 ml. of dimethyl sulfate was added. The mixture was heated under reflux for 1 hour, and then the benzene was removed by distillation. To the residue was added 300 ml. of 40% potassium hydroxide solution and the solution was heated for 5 hours on the steam-bath. The solution was cooled and extracted with ether. Distillation of the ether yielded 3.97 g. of residue. A refrigerated solution of the gummy base in ethanol yielded 1.81 g. of crystalline material; m.p. 92.5–94.5°. Recrystallization of this material from ethanol yielded 1.17 g. of *des*-dimethylapoerysotrine; m.p. 97.5–98°.

Anal. Calcd. for $C_{20}H_{23}NO_2$: C, 77.64; H, 7.49; N, 4.53. Found: C, 77.42; H, 7.66; N, 4.50.

The mother liquor from the above-described original crystalline material was concentrated to yield 1.34 g. of residue. A solution of this residue in 25 ml. of benzene was treated with 8 ml. of dimethyl sulfate and then refluxed for 2 hours. The benzene was distilled and the residue was dissolved in 100 ml. of 40% potassium hydroxide solution. After heating the aqueous solution for four hours on the steam-bath, it was cooled and extracted with ether. Removal of the ether left a gum which was dissolved in ethanol; crystals separated which melted at 130–131°. This compound might possibly be a *trans* form of *des*-dimethylapoerysotrine.

Anal. Calcd. for $C_{20}H_{23}NO_2$: C, 77.64; H, 7.49. Found: C, 77.52; H, 7.21.

Attempted Third Stage Hofmann Degradation on *des*-Dimethylapoerysotrine.—A 500-mg. sample of *des*-dimethylapoerysotrine, m.p. 97.5–98°, was dissolved in 20 ml. of benzene and 2 ml. of dimethyl sulfate was added. After heating on the steam-bath for 2 hours, the benzene was removed and the residue was dissolved in 40 ml. of 40% potassium hydroxide solution. When this solution was heated, the liberation of trimethylamine was not detected. The starting material, *des*-dimethylapoerysotrine, was recovered.

Hydrogenation of *des*-Dimethylapoerysotrine to Tetrahydro-*des*-dimethylapoerysotrine.—A 500-mg. sample of *des*-dimethylapoerysotrine, m.p. 97.5–98°, was dissolved in 125 ml. of ethanol and hydrogenated in the presence of Adams platinum catalyst. Two moles of hydrogen was absorbed. The yield of tetrahydro-*des*-dimethylapoerysotrine after crystallization was 473 mg., m.p. 58–59°. The compound was dried at 25° *in vacuo* before the analyses.

Anal. Calcd. for $C_{20}H_{27}NO_2$: C, 76.64; H, 8.68; N, 4.47; $2CH_3O$—, 19.80; $2CH_3N$=, 9.59; CH_3C≡, 4.80. Found: C, 77.11; H, 8.80; N, 4.68; CH_3O—, 18.94; CH_3N=, 11.50; CH_3C≡, 5.36.

One attempt to carry out an Emde degradation of this tetrahydro derivative failed to yield a nitrogen-free product.

N-Dimethylanthranilic Acid.—One gram of N-methylanthranilic acid (Eastman Kodak Co.) was dissolved in 25 ml. of methanol and an excess of diazomethane in ethereal solution was added. The mixture was heated on the steam-bath for about 30 minutes and the solvent was permitted to distil. A solution of the residue in 25 ml. of chloroform was washed with 5% sodium bicarbonate solution, and then evaporated. The solution of the residue in 10 ml. of 5% sodium hydroxide was heated at 80° for 40 minutes, cooled and extracted with chloroform. The alkaline solution was neutralized to *p*H 7 with hydrochloric acid, and extracted five times with chloroform; removal of the solvent gave 211 mg. of residue, which crystallized from ethyl ether. Two recrystallizations gave 66 mg. of N-dimethylanthranilic acid which melted constantly at 71–72°.

Indole from the Fusion of Erysodine with Potassium Hydroxide.—Twenty grams of erysodine was added in small portions to molten potassium hydroxide which was vigorously stirred mechanically. Reaction took place when each addition of erysodine was made. The heating and stirring was continued for 10 minutes after the last addition or until the frothing and rising of the melt had subsided. The cooled melt was treated as described for the fusion of erythraline,[2] and a yield of slightly more than 2 g. of indole was obtained; m.p. 52°.

Anal. Calcd. for C_8H_7N: C, 82.04; H, 6.02. Found: C, 81.93; H, 5.99.

4-Ethoxy-5-methoxyphthalic Anhydride from Erysodine.—To a solution of 1.087 g. of erysodine in 50 ml. of 5% potassium hydroxide solution, 10 ml. of freshly distilled diethyl sulfate was added and the mixture was shaken mechanically for 1.5 hours. To this alkaline solution, 300 ml. of 5% potassium permanganate solution was added dropwise with stirring. For the addition of the last 70 ml. of oxidant, the temperature was raised by a warm water-bath. The solution was cooled, acidified with hydrochloric acid, and gaseous sulfur dioxide was added until the manganese dioxide was gone. The clear solution was now extracted continuously with ether for 4 hours. Concentration of the ether yielded a residue which was dissolved in 10 ml. of 5% ammonium hydroxide. The solution was filtered and treated with a saturated solution of calcium chloride. The calcium oxalate was removed by filtration, and the filtrate was acidified with hydrochloric acid and extracted continuously with ether for 2 hours. Distillation of the ether left 171 mg. of yellow amorphous residue which was sublimed at 115° and 4×10^{-4} mm. The yellow crystalline sublimate melted at 184–186° and weighed 53 mg. Five recrystallizations of this material from anhydrous ethyl ether gave 4-ethoxy-5-methoxyphthalic anhydride, m.p. 192–193°. The melting point was constant and unchanged when a sample was mixed with a synthetic sample.

Anal. Calcd. for $C_{11}H_{10}O_5$: C, 59.48; H, 4.53. Found: C, 59.47; H, 4.82.

4-Ethoxy-5-methoxyphthalic Anhydride from Erysovine.—To a solution of 2.024 g. of erysovine in 100 ml. of 10% sodium hydroxide solution, 5 ml. of diethyl sulfate was added. The mixture was shaken mechanically for 1 hour. The solution was still alkaline, and a small amount of oil had separated. To this solution, 500 ml. of 5% potassium permanganate solution was added dropwise and with stirring. Sulfur dioxide was bubbled into the mixture until the manganese dioxide had disappeared. The solution was extracted continuously with ether for 6 hours, and concentration of the ether yielded 1.025 g. of residue. This material was dissolved in 50 ml. of 10% ammonium hydroxide and saturated calcium chloride solution was added until all of the oxalic acid was precipitated. The calcium oxalate was removed by filtration, and the filtrate was acidified with hydrochloric acid to *p*H 3.5. The solution was extracted continuously for 4 hours with ether; removal of the ether gave 357 mg. of a gummy solid. Sublimation of this material at 110–115° and 1.5×10^{-4} mm. gave a yellow sublimate; m.p. 189–191°. Resublimation gave 252 mg. of material; m.p. 191–192°. Recrystallization of this product from ethyl ether–petroleum ether gave 72 mg. of 4-ethoxy-5-methoxyphthalic anhydride; m.p. 192°. This melting point was unchanged when the product was mixed with a synthetic specimen.

Anal. Calcd. for $C_{11}H_{10}O_5$: C, 59.48; H, 4.53. Found: C, 59.32; H, 4.73.

4-Ethoxy-5-methoxyphthalic Anhydride and 4-Ethoxy-N-ethyl-5-methoxyphthalimide from "Erysocine."—The methylation and oxidation of 4 g. of "erysocine" (m.p. 162–163°, $[\alpha]^{25}D$ +233, in abs. alcohol) was carried out similar to the above-described experiments using erysovine and erysodine. The 4-ethoxy-5-methoxyphthalic anhydride melted at 192–193°. A 37-mg. portion of the anhydride was dissolved in ether and 2 ml. of anhydrous ethylamine was added. The mixture was concentrated to dryness, and the residue was heated at 105–110° bath temperature and 2 mm. until sublimation was complete; yield 38 mg., m.p. 198–199°. After two recrystallizations of this sublimate from methanol, the 4-ethoxy-N-ethyl-5-methoxyphthalimide melted at 202–203°.

Anal. Calcd. for $C_{13}N_{15}NO_4$: C, 62.65; H, 6.06. Found: C, 62.47; N, 5.94.

4-Ethoxy-5-methoxyphthalic Anhydride from Oxidation of 7-Ethoxy-6-methoxy-3,4-dihydroisoquinoline.[12]—Seven hundred milligrams of 7-ethoxy-6-methoxy-3,4-dihydroisoquinoline, m.p. 83–84°, was dissolved in 50 ml. of water and, while this solution was stirred mechanically, a solution of 2.3 g. of potassium permanganate in 200 ml. of water was added gradually. The mixture was warmed on the steam-bath to finish the oxidation. The solution was acidified with hydrochloric acid and treated with sulfur dioxide. When the manganese dioxide was gone, the solution was extracted continuously with ethyl ether for 12 hours. The ether extract yielded 291 mg. of residue which was dissolved in 30 ml. of 5% ammonium hydroxide. Calcium chloride solution was added until precipitation was complete. The filtrate from the precipitate was acidified and extracted continuously with ether for 2 hours. The ether residue weighed 247 mg. It was sublimed at 7×10^{-4} mm. and 125–130° bath temperature. The yellow sublimate weighed 206 mg. and melted at 187–189°. After two recrystallizations from ether, the 4-ethoxy-5-methoxyphthalic anhydride melted constantly at 192–193°.

Anal. Calcd. for $C_{11}H_{10}O_5$: C, 59.48; H, 4.53. Found: C, 59.55; H, 4.52.

Summary

Erysopine and erysovine have been converted into apoerysopine by reaction with hydrobromic acid. Hofmann degradation of apoerysopine using dimethyl sulfate and alkali has yielded *des*-dimethylapoerysotrine after two stages. *des*-Dimethylapoerysotrine and tetrahydro-*des*-dimethylapoerysotrine were not degraded to nitrogen-free products. Alkali fusion of erysodine gave indole. Ethylation and oxidation of erysodine, erysovine and "erysocine" gave 4-ethoxy-5-methoxyphthalic anhydride. This phthalic derivative was also obtained from 7-ethoxy-6-methoxy-3,4-dihydroisoquinoline.

It was found that erythraline has the same ring system as erysopine and related erysoalkaloids by degradation of erythraline to apoerysopine.

Interpretation of these reactions and products permits structural formulations of erysopine, erysodine and erysovine, erythraline and other related alkaloids.

(12) E. Späth and A. Dobrowsky, *Ber.*, **58**, 1274 (1925).

RAHWAY, NEW JERSEY RECEIVED JUNE 2, 1950

[CONTRIBUTION FROM THE DIVISION OF PLANT BIOCHEMISTRY, COLLEGE OF AGRICULTURE, UNIVERSITY OF CALIFORNIA]

Starch. IV. The Molecular Constitution of Amylose Subfractions

BY A. L. POTTER AND W. Z. HASSID

If the amylose component of starch consists of unbranched chains, as commonly assumed, its molecular weight determined by the periodate oxidation end-group method should be equal to that estimated by osmotic pressure measurements. However, analytical results are not always in accord with this assumption. Examination of the data obtained for amyloses of seven different sources[1] showed that three of them, potato, Easter lily and apple, had essentially the same molecular weights by the two methods, indicating that these amyloses are apparently unbranched. The other four amyloses were found to possess lower degrees of polymerization when determined by the periodate oxidation method than by osmotic pressure measurements. These results imply the possible existence of branching in some amyloses.

In order to obtain further information regarding the question of branching in amylose, a number of subfractions from potato and corn amyloses were studied. The amylose subfractions were contributed by T. J. Schoch of the Corn Products Refining Company. Their method of preparation is given by Lansky, Kooi and Schoch.[2] The methods employed for the determination of end-groups, osmotic pressure and viscosity measurements are described in the previous papers.[1]

Comparison of the data obtained from the two methods showed that the molecular weights of the potato subfractions (Table I, Potato, P-7/9-A, 17a and 17f) and all of the corn amylose subfractions (C-148/150-A, 14a, 14b, 14c and 13c) were significantly higher when determined by osmotic pressure measurements than when estimated by the periodate oxidation method. Their average number of non-reducing terminal glucose units per molecule was considerably greater than 1.0.

TABLE I

COMPARISON OF DEGREE OF POLYMERIZATION OF AMYLOSE SUBFRACTIONS OBTAINED BY OSMOTIC PRESSURE MEASUREMENTS AND PERIODATE OXIDATION END-GROUP METHOD

Source	Subfraction	No additional *n*-butanol repptns.	DP, osmotic pressure	DP, periodate oxidation	Average no. of non-reducing terminal glucose units per molecule
Potato	P-7/9-A, 17a		1600	1320	1.6
Potato	P-7/9-A, 17a	2	1600	1540	1.1
Potato	P-7/9-A, 17b		1230	1140	1.2
Potato	P-7/9-A, 17c		970	1000	0.9
Potato	P-7/9-A, 17d		900	940	0.9
Potato	P-7/9-A, 17e		890	900	1.0
Potato	P-7/9-A, 17f		930	630	2.4
Potato	P-7/9-A, 17f	2	880	880	1.0
Corn	C-148/150-A, 14a		1150	380	7.2
Corn	C-148/150-A, 14a	2	1150	450	5.6
Corn	C-148/150-A, 14b		890	360	5.4
Corn	C-148/150-A, 14b	2	890	510	3.2
Corn	C-148/150-A, 14b	3	890	510	3.2
Corn	C-148/150-A, 14c		670	470	2.3
Corn	C-148/150-A, 14c	2	670	540	1.7
Corn	C-148/150-A, 13c		560	340	2.9

If these discrepancies were due to the presence of amylopectin impurities, it should be possible to eliminate the latter by repeated recrystallization of the amyloses with *n*-butanol. In order to check

(1) (a) A. L. Potter and W. Z. Hassid, THIS JOURNAL, **70**, 3488 (1948); (b) **70**, 3774 (1948); (c) A. L. Potter, W. Z. Hassid and M. A. Joslyn, *ibid.*, **71**, 4075 (1949).

(2) S. Lansky, M. Kooi and T. J. Schoch, *ibid.*, **71**, 4066 (1949).